

In the Specification:

Please amend the specification as shown:

Please delete the paragraph on page 1, lines 3-7, and replace it with the following paragraph:

This is a Continuation in part of United States Application (Serial Number ~~not yet assigned~~
10/650,277) filed August 28, 2003 entitled "METHOD OF RESISTING OSTEOCLAST FORMATION"

which was a traditional application claiming priority from United States provisional application Serial Number 60/407,335 filed August 30, 2002 entitled "METHOD OF RESISTING OSTEOCLAST FORMATION".

Please delete the paragraph on page 9, line 20, to page 10, line 3, and replace it with the following paragraph:

The full-length ECF-L cDNA was generated by PCR using the mouse OCL cDNA as a template and specific primers sets for mECF-L (5'-ACACCATGGCCAAGCTCATT-3' (sense) (SEQ ID NO: 1) and 5'-TGCAGAATGCGCTGTGGAAA-3' (antisense) (SEQ ID NO: 2)). The PCR conditions were 94°C for 30 sec, 60°C for 30 sec, 72°C for 2 min and 40 cycles. The PCR product was subcloned into the TA vector and sequenced. The cDNA was digested with *Eco*RI and cloned into the mammalian expression vector pcDNA3 (Invitrogen) and transfected into 293 cells to express mECF-L.

Please delete the paragraph on page 11, line 21, to page 12, line 6, and replace it with the following paragraph:

To determine if native ECF-L was involved in OCL formation, we designed AS and SS S-oligonucleotides (5'-AAGAATGAGCTTGGCCATGGTGCTTCACG-3' (SEQ ID NO: 3) and 5'-CGTGAAGACACCATGGCCAAGCTCATTCTT-3' (SEQ ID NO: 4)) that included the ATG and ribosome binding site of the mECF-L gene. The AS and SS oligonucleotides were added at varying

concentrations to mouse bone marrow cultures stimulated with 10^{-9} M 1,25-(OH)₂D₃. Every 2 days, half of the media was replaced with the fresh media containing the SS-oligonucleotide and 10^{-9} M 1,25-(OH)₂D₃. At the end of the culture periods, the cells were fixed and stained for TRAP activity, and the number of TRAP(+) MNCs was determined.

Please delete the paragraph on page 13, lines 5-17, and replace it with the following paragraph:

Recombinant mECF-L (rmECF-L) was expressed in the BL21 E. coli strain using the pET14b expression vector system (Novagen, Inc., Madison, WI) according to the manufacturer's protocol. The nucleotide sequence encoding the mECF-L cDNA was amplified by PCR with sense primers (5'-CGAGGATCCGATGGCCAAGCTCATTCTGTC-3') (SEQ ID NO: 5) and antisense primers (5'-CGAGGATCCTCAATAAGGGCCCTTGCAACT-3') (SEQ ID NO: 6) (underlined sequences represent *Bam*HI site.) The PCR product was digested with *Bam*HI site and then cloned into the pET 14b vector in frame with the 6x His tag (SEQ ID NO: 7). The plasmid construct was transformed into the BL21 (DE3) E.coli, and the recombinant ECF-L was induced by treatment with 0.5 mM IPTG for 4 hours. The cells were pelleted by centrifugation, washed with PBS and resuspended in His buffer containing 8 M urea. After sonication and centrifugation, the supernatant was loaded onto Ni-NTA Superflow bulk resin (Qiagen, Valencia, CA, USA) and the 6xHis-r ECF-L fusion protein (6xHis tag shown in SEQ ID NO: 7) was eluted with a 50 – 100 mM imidazole gradient. The eluent was dialyzed against milli Q water and injected into rabbit to generate the anti ECF-L polyclonal sera.

Please delete the paragraph on page 15, lines 2-9, and replace it with the following paragraph:

ECF-L cDNA was generated by PCR using T7 and antisense primer (5'-ATCGTAATCCATAAGGGCCCTTGCAACTG-3') (SEQ ID NO: 8), and the EcoRV-digested PCR-product was fused with the Fc coding domain of human IgG1. The mECF-L-Fc construct was stably transfected into 293 cells using a CaPO₄ mammalian transfection kit (Stratagene, La Jolla, CA, USA) according to the manufacturer's protocol. One hundred microgram of mECF-L-Fc fusion

protein was purified from 1 L of 293 cells conditioned media by protein G affinity chromatography (Roche Diagnostics). The effects of purified mECF-L-Fc fusion protein on OCL formation/activation was tested in murine bone marrow cultures as described above.

Please delete the paragraph on page 15, lines 13-20, and replace it with the following paragraph:

Mouse bone marrow cells (1.2×10^7 /well) were cultured with mECF-L conditioned media in 6-well plates for 2 days. Total RNA was extracted with RNA-BEE (Tel Test, Friendswood, TX) according to the manufacturer's protocol, and the expression levels of mouse RANKL mRNA were determined by RT-PCR analysis. The PCR conditions were 94°C for 30 sec, 58°C for 30 sec, 72°C for 1 min and 28 cycles. PCR primer sequences for mouse RANKL are as follows: sense primers, 5'-GAAGGTACTCGTAGCTAAGG -3' (sense) (SEQ ID NO: 9) and 5'-GGCTATGTCAGCTCCTAAAG-3' (antisense) (SEQ ID NO: 10). GAPDH was used as an internal control using primer sequences 5'-ACCACAGTCCATGCCATCAC -3' (sense) (SEQ ID NO: 11) and 5'-TCCACCACCCTGTTGCTGTA-3' (antisense) (SEQ ID NO: 12).